

ONDANA RECH CO.1. John Skock College Il15 Cas9-CKO Strategy Ronald Colons

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Project Overview



Project Name Il15

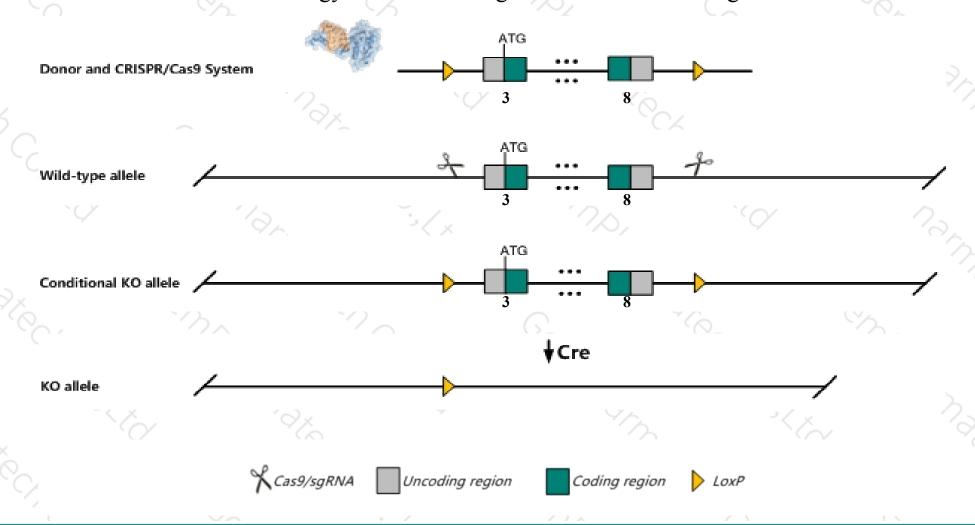
Project type Cas9-CKO

Strain background C57BL/6J

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Il15* gene. The schematic diagram is as follows:



Technical routes



- The *Il15* gene has 10 transcripts. According to the structure of *Il15* gene, exon3-exon8 of *Il15-201* (ENSMUST00000034148.6) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Il15* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA and Donor were microinjected into the fertilized eggs of C57BL/6J mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6J mice.
- > The flox mice was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

Notice



- ➤ According to the existing MGI data, Mice homozygous for disruptions of this gene have normal life spans but display a variety of immune system abnormalities and maternal placental defects.
- The *Il15* gene is located on the Chr8. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- > This Strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)



II15 interleukin 15 [Mus musculus (house mouse)]

Gene ID: 16168, updated on 19-Mar-2019

Summary

Official Symbol II15 provided by MGI

Official Full Name interleukin 15 provided by MGI

Primary source MGI:MGI:103014

See related Ensembl:ENSMUSG00000031712

Gene type protein coding RefSeq status REVIEWED

Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;

Muroidea; Muridae; Murinae; Mus; Mus

Also known as Al503618, IL-15

Summary This gene encodes a a pleiotropic cytokine of the interleukin family of proteins that plays important roles in the innate and adaptive cell

homeostasis, as well as peripheral immune function. The encoded protein undergoes proteolytic processing to generate a mature cytokine that stimulates the proliferation of natural killer cells. The transgenic mice overexpressing the encoded protein exhibit an increase in the number of memory CD8+ T cells in a naive state and enhanced protection against bacterial infections. Mice lacking the encoded protein

exhibit impaired protection against a strain of attenuated Mycobacterium. [provided by RefSeq, Aug 2016]

Expression Broad expression in heart adult (RPKM 5.2), duodenum adult (RPKM 3.3) and 24 other tissuesSee more

Orthologs <u>human</u> all

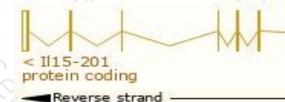
Transcript information (Ensembl)



The gene has 10 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
II15-201	ENSMUST00000034148.6	1908	<u>162aa</u>	Protein coding	CCDS40401	P48346 Q3U1Z6	TSL:1 GENCODE basic APPRIS P1
II15-202	ENSMUST00000209363.1	1251	<u>162aa</u>	Protein coding	CCDS40401	P48346 Q3U1Z6	TSL:1 GENCODE basic APPRIS P1
II15-203	ENSMUST00000209573.2	1164	<u>162aa</u>	Protein coding	CCDS40401	P48346 Q3U1Z6	TSL:5 GENCODE basic APPRIS P1
II15-205	ENSMUST00000210094.1	430	<u>48aa</u>	Protein coding	-	A0A1B0GQZ2	CDS 3' incomplete TSL:3
II15-209	ENSMUST00000211565.1	766	<u>61aa</u>	Nonsense mediated decay	-	A0A1B0GQZ8	TSL:3
II15-206	ENSMUST00000210472.1	618	No protein	Processed transcript	-	-	TSL:3
II15-210	ENSMUST00000211722.1	433	No protein	Processed transcript	-	-	TSL:1
II15-204	ENSMUST00000209687.1	430	No protein	Processed transcript	-	-	TSL:1
II15-208	ENSMUST00000211282.1	258	No protein	Processed transcript	-	-	TSL:5
II15-207	ENSMUST00000210885.1	234	No protein	Processed transcript	-	-	TSL:5

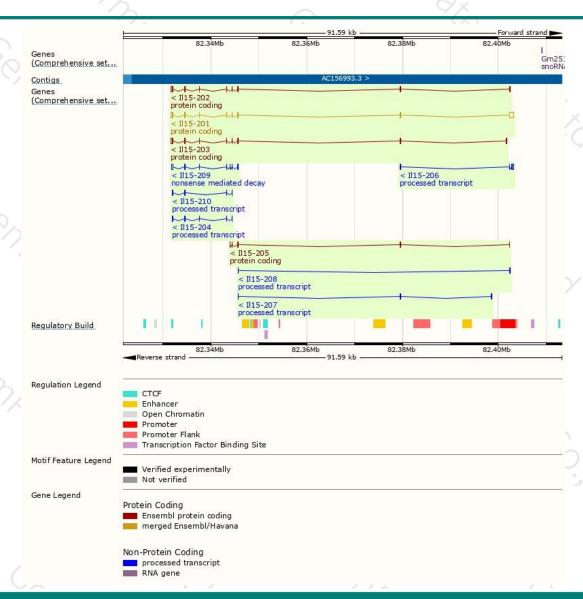
The strategy is based on the design of *Il15-201* transcript, The transcription is shown below



71.59 kb

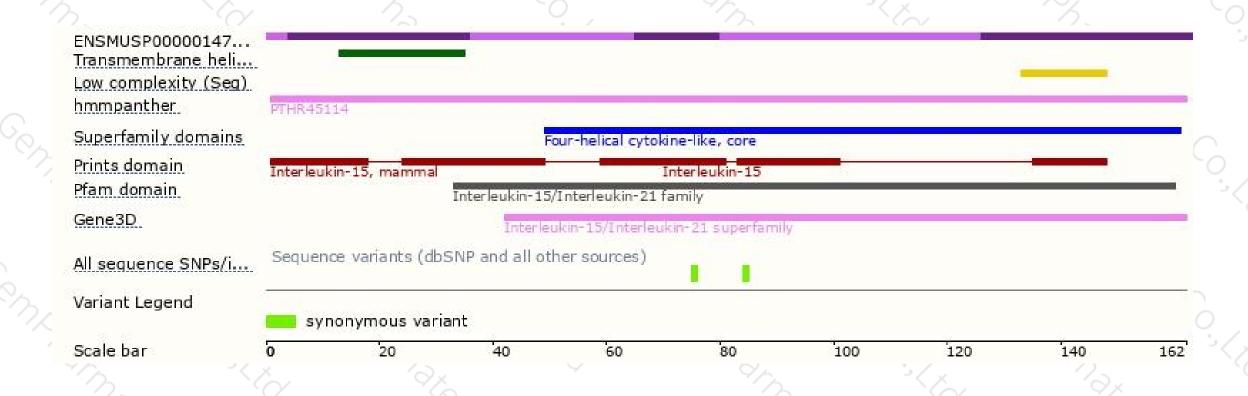
Genomic location distribution





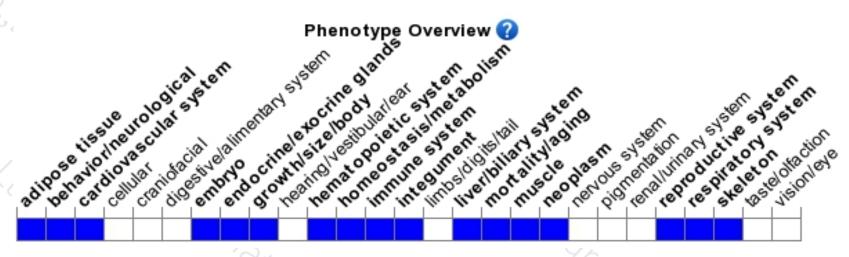
Protein domain





Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data, Mice homozygous for disruptions of this gene have normal life spans but display a variety of immune system abnormalities and maternal placental defects.



If you have any questions, you are welcome to inquire.

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